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Two new records of *Mucorales* from the Brazilian semi-arid region

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Abstract — *Apophysomyces elegans* and *Mycotypha microspora* are recorded for the first time in Brazil based on isolates from semiarid soil in the Northeast part of the country.

Key words — *Zygomycetes*, *Mucoromycotina*, taxonomy

Introduction

Apophysomyces and *Mycotypha* belong to the subphylum *Mucoromycotina* (Hibbett et al. 2007), family *Mucoraceae*, order *Mucorales* (Benny 2005). *Apophysomyces* was first described by Misra et al. (1979), and the description of *A. elegans* (the monotype) was based on two specimens isolated from soil. This species typically produces a pyriform, apophysate multispored sporangia developed on a sporangiophore with a funnel-shaped to bell-shaped apophysis. *Apophysomyces elegans* has also been reported as an agent of zygomycosis in immunocompromised patients (Kimura et al. 1999; Liang et al. 2006; Chakrabarti et al. 2008; Reddy et al. 2008).

Mycotypha was introduced by Fenner (1932), who described a single species, *M. microspora*. Six species have been included in the genus, but Benny et al. (1985) accepted only three as true *Mycotypha* species. *Mycotypha microspora* was isolated as a contaminant on a plate culture of a pathogen of bitter orange (*Citrus aurantium*) and was first classified in *Mucoraceae*. Since then, *Mycotypha* has been placed in the *Choanephoraceae* (Bessey 1950) and the *Cunninghamellaceae* (Hesseltine 1952). Novak & Backus (1963) described *M. africana*, which produces zygospores with a typically mucoraceous form. Young (1969), based on the electron and phase-contrast microscopy of spores, reported that *Mycotypha* should be included in the *Thamnidiaceae*. Later,

Benny et al. (1985) proposed the family *Mycotyphaceae*, including a new species *M. indica*. *Mycotypha microspora* is characterized by sporophores terminating in a mostly cylindrical fertile vesicle bearing dimorphic sporangiola subtended by conical denticles. Yeast-like budding cells and thin-walled chlamydospores are also characteristics of this species.

The purpose of this manuscript is to report the first occurrence of *Mycotypha microspora* and *Apophysomyces elegans* in Brazil. For *M. microspora* this also represents the first record for South America.

Materials and methods

Soil samples were collected at Belém de São Francisco (8°33'59"S, 38°49'59"W) and Triunfo (7°52'28"S, 38°06'03"W), located in the semi-arid region of the State of Pernambuco, Northeastern Brazil. Belém de São Francisco is characterized by xerophilous vegetation with patches of deciduous forest. The typical biome is named caatinga and the climate is tropical semi-arid. Triunfo comprises semi deciduous forest and, according to Koeppen's classification, the climate is hot and humid tropical. Both areas are included in the Brazilian semi-arid region, which covers more than 969,589 km^2 (Ministério da Integração Nacional 2005).

The samples of soil were collected with a sterilized spatula, placed in plastic bags and taken to the laboratory. Soil particles (5 mg) were placed on sets of Petri dishes containing MEYE (Benny 2008) plus chloramphenicol (100mg/L). The plates were left on a bench at room temperature (28 ± 2°C) under light and dark periods for 72 hours. Fragments of mycelium were removed directly from the samples at the stereomicroscope and transferred to Petri dishes with M agar (O'Donnell 1979). Identification and descriptions were based on macroscopical (color, aspect and diameter of the colonies) and microscopical (microstructures) characters according to Benny & Benjamin (1976) and Misra et al. (1979).

Taxonomy

Apophysomyces elegans P.C. Misra, K.J. Srivast. & Lata, Mycotaxon 8(2): 377 (1979)
Fig. 1 a–d

Specimen examined: Brazil, Pernambuco, Triunfo, soil, Jan. 2010, A.L.C.M.A. Santiago (URM-Culture collection 6169).

Colonies remaining white on M agar, reverse pale yellow, 9 cm diam in 72 hour at 28°C. Sporangiophores growing slowly, after 7 days, often single, developing at right angles from aerial stolon-like hyphae which generally becomes delimited by two septa near the place of origin of the sporangiophore; erect, unbranched, thick-walled, smooth, light brown, becoming darker near the base and darker and thicker below the apophyses, up to 550 µm long and 5 µm wide near the base. Sporangia hyaline at first, becoming light yellowish-brown, terminally, pyriform, distinctly apophysate, 20–50 µm diam. Apophyses funnel-shaped to bell-shaped, 12–47 µm high and 17–27.5 µm diam at the

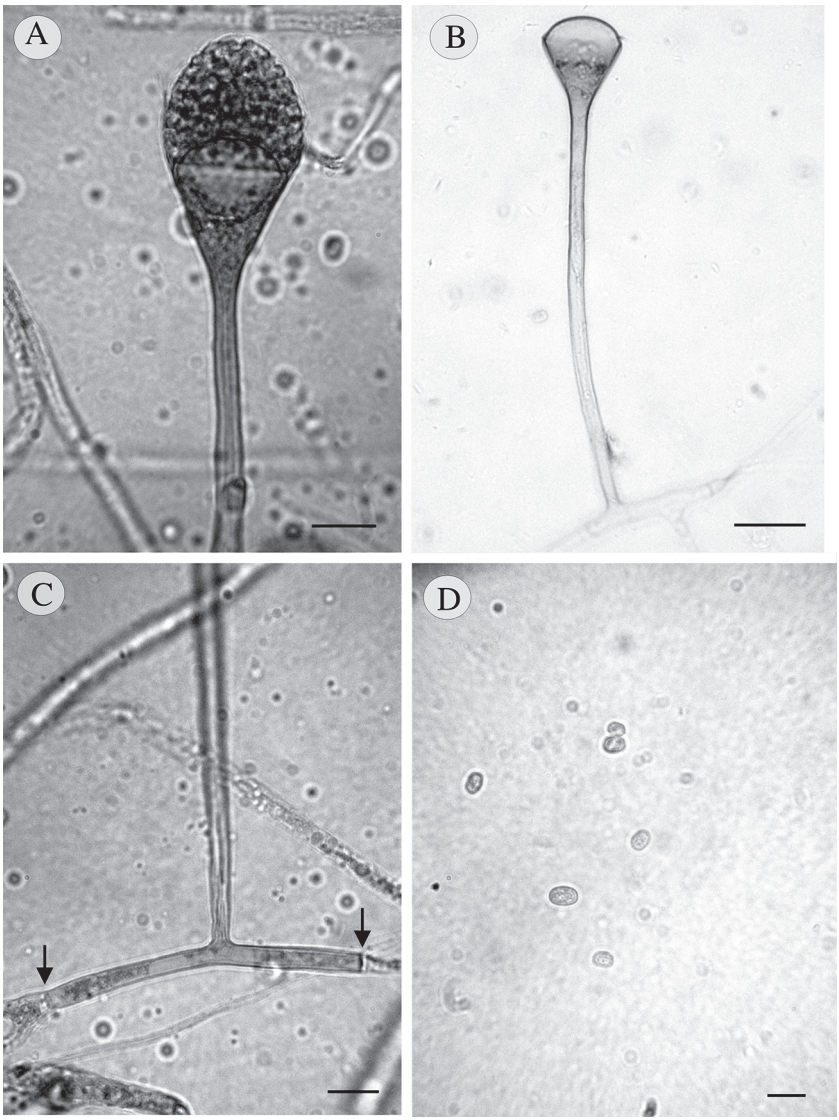

Fig. 1 *Apophysomyces elegans*. A) Sporangiophore with sporangium; B) Sporangiophore with funnel-shaped apophysis and columella; C) Stolon-like hypha delimited by two septa near the place of origin of the sporangiophore; D) Sporangiospores. Scale bars: A, C, D = 10μm, B = 20 μm.

widest part; smooth-walled, light brownish. Columellae hemispherical, thin-walled, subhyaline, 20–30 μm diam, collar distinct. Sporangiospores oblong,

sometimes subglobose, subhyaline, minutely roughened, 4.5–8.5(–12.5) × 4–5.5(–6.5) µm. Rhizoids unbranched, subhyaline. Zygospores not observed.

Habitat: Soil

Geographic distribution: Australia (Cooter et al. 1990), Caribbean (Meis et al. 1994), Colombia (Ruiz et al. 2004), India (Mirza et al. 1979; Lakshmi et al. 1993; Shakrabarti el al. 2003) and USA (Blair et al. 2002; Liang et al. 2006; Ferguson et al. 2007).

Remarks: The characteristics of the *Apophysomyces elegans* strains reported here show a close similarity with the original description of Misra et al. (1979). However, differences in colony color and sporangiospore walls were observed. The colonies were persistently white, as also described by Lakshmi et al. (1993), but Misra et al. (1979) and Cooter et al. (1990) reported colonies as white at first, becoming brownish-gray, and then creamy white to buff with age. Recently, Reedy et al. (2008) described the colonies as initially white, turning brownish-gray or yellow. The fact that different authors have used dissimilar culture media for descriptions may explain this variation of color. Curiously, the *A. elegans* sporangiospores described here are minutely roughened, differing from the smooth ones reported by Misra et al. (1979). However, we did not consider these differences enough to characterize a new taxon. *Apophysomyces elegans* has some microscopic features similar to those of species of *Absidia*, like sporangiophores arising from stolons and pyriform, apophysate sporangia. Nevertheless, *Apophysomyces* differs from *Absidia* in bearing a more pronounced, funnel-shaped to bell-shaped, apophysis. In addition, the sporangiophore wall below the apophyses is dark and thick in *Apophysomyces* (Mirza 1979; Lakshmi 1993).

Mycotypha microspora Fenner, Mycologia 24: 196 (1932) Fig. 2 a–d

Specimen examined: Brazil, Pernambuco, Belém de São Francisco, soil, Jan. 2010, A. L. C. M. A. Santiago (URM-Culture collection 6170).

Colony with limited growth after 15 days at 28°C in M agar; more or less zonate, later deep gray or brown with age. Sporophores simple at first, some secondarily branched, hyaline at first, becoming grayish brown in age, irregularly multiseptate, particularly below the vesicle, 3 mm high, 3–18.5 µm diam. Fertile Vesicles terminal, mostly cylindrical, rounded at the apex, appearing minutely roughened, bearing sporangiola over entire surface, except at extreme tip, 20–580 × 10–40 µm. Sporangiola dimorphic, forming two different layers over surface of vesicle; at outer layer, ovoid to obovoid, 4–6 × 3–5 µm, pale bluish-gray, smooth, globose to subglobose borne on conical pedicels; at inner layer, 3–5.5 µm in diam, pale bluish-gray, smooth, born on conical pedicels. After dehiscence, the sporangioles bear remnant of pedicel. Zygospores not observed.

Habitat: Soil

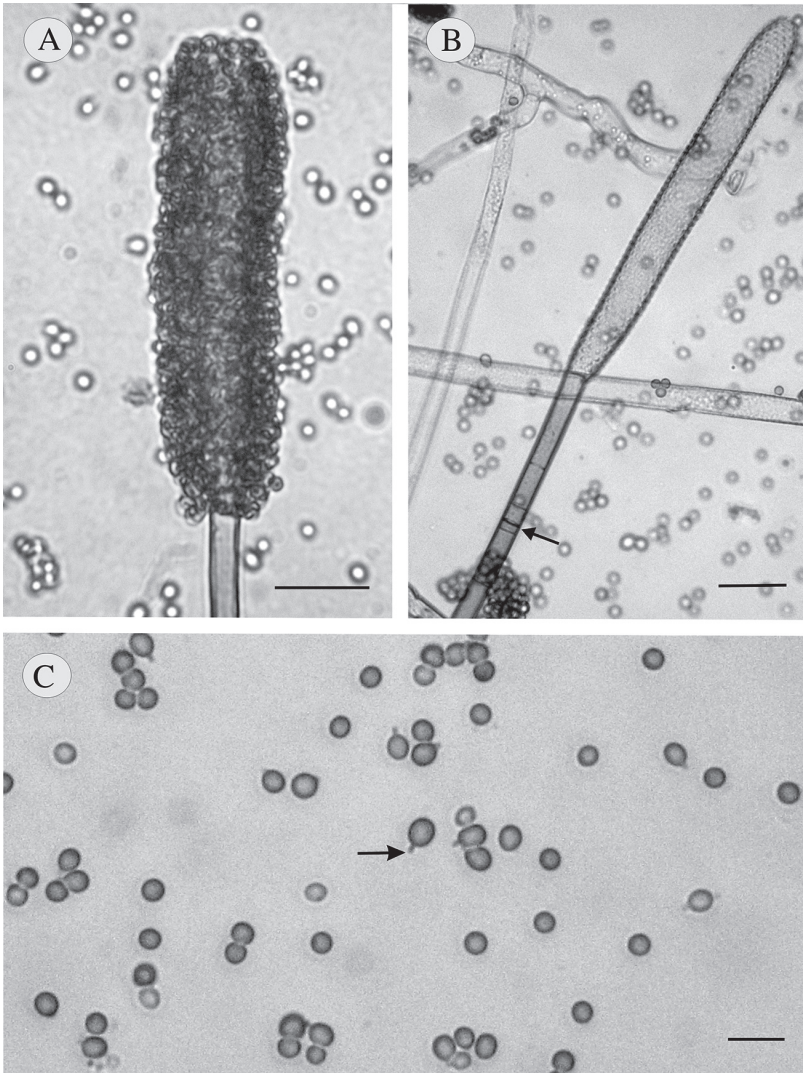

Fig. 2 *Mycotypha microspora.* A) Sporangiophore with terminal fertile vesicle and sporangiola; B) Terminal fertile vesicle after dehiscence of the sporangiola; septa produced near the vesicle. C) Globose and ovoid to obovoid sporangiola with remnant of pedicels.
Scale bars: A, B = 30 µm; C = 10 µm.

Geographic distribution: Belgium (IHEM), Finland (IMI), France (Lacroix et al. 2007; IHEM), Germany (IMI), Great Britain (IMI), India (Ray & Mukerji 1961), Japan (NBRC), Libya (IMI), Netherlands (CBS), Nigeria (IMI), Poland (IMI), USSR (CBS), Thailand (CBS), Turkey (MUCL), USA (Benny & Benjamin 1976).

Remarks: The strain characteristics of *M. microspora* reported here are very close to the original description by Benny & Benjamin (1976). The known species of *Mycotypha* are morphologically similar, but *M. microspora* differs from *M. africana* in producing ovoid to obovoid external sporangiola, while in the latter the external sporangiola are cylindrical. In *M. microspora* the septa in the sporophore are usually produced near the apex but may also be formed near the base, while in *M. indica* the septa are only produced near the base (Benny et al. 1985).

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